

## The morphological characteristics of the sparganum stage of the Malaysian *Spirometra* species

Kavana N J<sup>1</sup>, Lim L H S<sup>2</sup>, Ambu S<sup>3</sup>

**Background:** The present study describes the morphology of sparganum (larva) of the Malaysian *Spirometra* spp. collected from naturally infected frogs (*Rana cancrivora*) from rice fields in Tanjung Karang, Malaysia.

**Materials and Methods:** Spargana of *Spirometra* spp. collected from naturally infected frogs (*Rana cancrivora*) were used for the morphological studies. Stretched on a metal ruler, measurements of the worm were recorded. Specimens were stained in Alum-carmin.

**Results:** The length of the body ranged from 11-50 mm and the width ranged from 0.5-1.5 mm. Specimens stained with Alum-carmin showed ridges (formation of segments) on the surface of the body, and no sexual organs in the body.

**Conclusion:** The Malaysian *Spirometra* spp. are similar in measurement and morphology to *Spirometra erinacei* but further studies are required for confirmation.

IeJSME 2015 9(2): 22-24

*Key Words:* *Spirometra*, *spargana*, *scolex*, *body*, *morphology*

### Introduction

*Spirometra* spp. is a cestode and its larval form is known as the spargana. This larval form causes larval migrans in humans who consume under-cooked frog meat. There are 5 stages in the life cycle of *Spirometra* spp., which include the egg, coracidium, proceroid, plerocercoid (spargana) and the adult worm. The infective stages are the coracidium, proceroid and spargana. Egg and adult stages are not infective. The coracidium is infective to the cyclops (a zooplankton) which is the first intermediate host.<sup>1</sup> The proceroid is infective to the second intermediate hosts which are amphibians, reptiles, birds and mammals.<sup>2,3,4</sup> The spargana infect the definitive hosts (Canidae and Felidae) when they eat the second intermediate hosts in which they develop to the

adult stage in their intestines. There is a difference in the morphology of every stage and which clearly distinguishes one stage from the other. The infection with spargana in humans is known as sparganosis.<sup>5,6</sup> In this paper we highlight some morphological characteristics of the plerocercoid (spargana) stage of the Malaysian *Spirometra* spp.

### Materials and Methods

Spargana were collected from naturally infected frogs belonging to the species *Rana cancrivora*. Spargana were collected from 211 naturally infected frogs and 13 of these specimens were used for the measurements. The worms were placed in petri dishes with normal saline for 5 minutes to relax. They were then picked with fine forceps, stretched on a metal ruler and the measurements recorded. After this the spargana were killed with hot 10% formal saline and fixed with 70% ethanol. The specimens were hydrated by transferring them to down-graded alcohol series 70%, 50%, and 30% for 10 minutes each. After this the specimens were placed in distilled water for 10 minutes and then stained in Alum-carmin overnight and destained with dilute hydrochloric acid (HCl). They were then transferred to distilled water for 10 minutes and dehydrated in an upgraded alcohol series of 30%, 50%, 70%, 95%, and 100% with 10 minutes intervals. Thereafter the specimens were cleared in xylene for 2 minutes and mounted on glass slides using Canada balsam. The slides were incubated in the oven at 60°C for one week. The mounted specimens were examined under light microscopy (Leitz Wetzlar).

### Results

The relaxed whole worms collected from the frogs are shown in Fig. 1. The spargana are long, have a large head, slender body and are ribbon-like in appearance. The length of the body ranges from 11-50 mm and the width from 0.5-1.5 mm as shown in Table 1. The scolex has a depression which is the undeveloped bothrium seen on the anterior aspect of the worm (Fig. 2).

<sup>1</sup>Institute of Biological Sciences, University of Malaya, Kuala Lumpur, MALAYSIA. (Currently at the Institute of Development Studies, Mzumbe University, P.O. BOX 83, Mzumbe, Morogoro, TANZANIA)

<sup>2</sup>Institute of Biological Sciences, University of Malaya, Kuala Lumpur, MALAYSIA

<sup>3</sup>Institute for Research Development and Innovation, International Medical University, Kuala Lumpur, MALAYSIA

Address for Correspondence:

Prof Stephen Ambu, Institute for Research Development and Innovation, International Medical University, Kuala Lumpur, MALAYSIA

Email: Stephen\_ambu@imu.edu.my

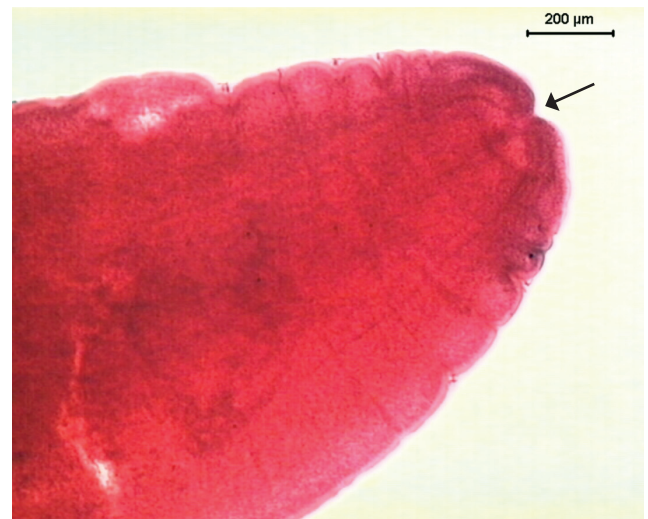
The body has ridges on the surface with no sexual organs seen in the body segments (Fig. 3).

**Table 1: Measurements of 13 spargana recovered from the naturally infected frogs (*R. cancrivora*) collected from Tanjung Karang, Malaysia.**

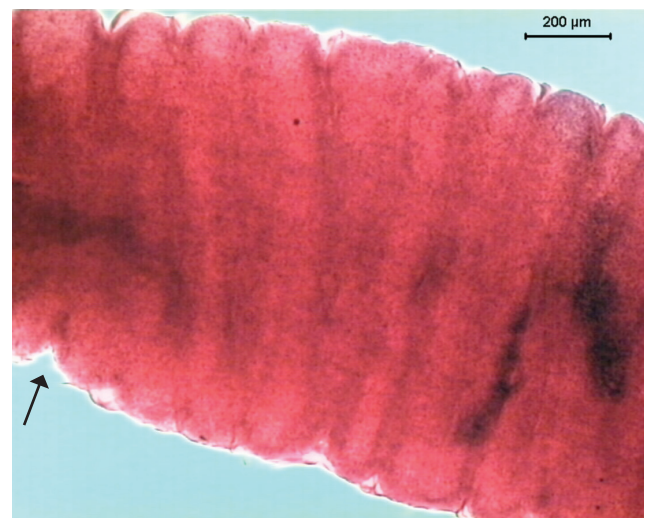
NO. OF SPARGANA	LENGTH (MM)	WIDTH (MM)
1	44	1
2	35	0.75
3	50	1
4	29	1
5	25	1.5
6	21	0.5
7	35	1
8	14	1
9	50	1
10	12	1
11	11	0.5
12	17	0.5
13	16	0.5
<b>Mean</b>	<b>27.6</b>	<b>0.9</b>
<b>Range</b>	<b>11 - 50</b>	<b>0.5 -1.5</b>



**Fig.1: Spargana recovered from naturally infected frogs (*R. cancrivora*) shown to scale.**



**Fig.2: Scolex of the sparganum of the Malaysian *Spirometra* spp. showing the under-developed bothrium (depression) on the anterior end (arrow). (Alum-carmin stain)**



**Fig.3: The sparganum collected from the naturally infected frogs (*R. cancrivora*) showing early segmentation marked by ridges (arrow). No sexual organs are seen in the body cavity. (Alum-carmin stain).**

## Discussion

Sparganum, the larval form of *Spirometra* spp. lacks specificity for intermediate hosts. It infects most vertebrates except fishes and are highly paratenic.<sup>7</sup> Once ingested by the host it penetrates the intestine and reestablishes itself in the tissues of the body wall or under the skin. Spargana can survive in the intermediate and paratenic hosts for a long time until they are eaten by the definitive host.<sup>8</sup> During their development in the second intermediate hosts the larvae grow slowly and increase in length and width.<sup>9</sup>

The spargana in the present study have a club shaped anterior scolex, with a broad middle part as indicated by ridges (body) with no sexual organs and a slender posterior part. This conforms to the description of the spargana of the *Spirometra* spp.<sup>10,11,12</sup> However more work has to be carried out on these larval forms to confirm the species of the *Spirometra* spp. found in these wild caught frogs in Tanjung Karang, Malaysia.

## Acknowledgement

This study is a part of the thesis submitted to the University of Malaya in fulfillment for an MSc degree by N.J. Kavana. We wish to acknowledge the Commonwealth Secretariat, London for the fellowship and University of Malaya, Malaysia for the financial support of Vote F. Appreciation is also extended to all other staff who provided the technical support to carry out these studies at the University.

## REFERENCES

1. Li CH, Faust EC. Infection of cyclops with the coracidium of oriental diphyllbothrids and their development to the mature proceroid stage. Proc. Soc. Exp. Biol. Med. 1928; 26: 250-1.
2. Okumura, (1919). The life history of *Sparganum mansoni*. Jap. Med. World. 1919; 1: 190-7.
3. Lee SY, We JS, Bae J, Seo BS. (1975). Some aspects of human sparganosis in Korea. Korean J Parasit. 1975; 13: 60-77.
4. Beaver PC, Jung RC, Cupp EW. Clinical Parasitology. 1984; 825. 9<sup>th</sup> edition, Philadelphia, Lea & Febiger.
5. Wardle RA, McLeod JA. The Zoology of Tapeworms. Minneapolis, University of Minnesota Press.1952; 780 Sparks AK, Naefie RC, Connor DH. Sparganosis. In Pathology of Tropical and Extraordinary Diseases. Editors. C.H. Binford, Institute of Pathology, Washington. 1976.
6. Mueller JF. Host-parasite relationships as illustrated by the cestode *Spirometra mansonioides*. Proc. 26<sup>th</sup> Annual Biol. Colloq. Univ. of Oregon, 1966: 15-58.
7. Lee SH, We JS, Sohn WM, Hong ST, Chai JY. Experimental life history of *Spirometra erinacei*. Korean J Parasitol 1990; 28 (3): 161-73.
8. Hong ST, Kim KJ, Huh S, Lee YS, Chai JY, Lee SH. The changes of histopathology and serum anti-sparganum IgG in experimental sparganosis of mice. Korean J Parasit. 1989; 27 (4): 261-9.
9. Kavana NJ, Lim LHS, Ambu S. The life-cycle of *Spirometra* species from Peninsular Malaysia. Trop Biomed 2014; 31: 487-95.
10. Mueller, J F 1938. Studies on *Sparganum mansonioides* and *Sparganum proliferum*. American Journal of Tropical Medicine.18: 303-28
11. Iwata, S (1972). Experimental and morphological studies of Manson's tapeworm *Diphyllobotrium erinacei*, Rudolphi. Special reference with its scientific name and relationship with *Sparganum proliferum*. Ijima Progress of Medical Parasitology in Japan 4: 536-90.
12. Miyazaki, I. Helminthic Zoonoses. An illustrated book of helminthic zoonoses. International Medical Foundation Japan, Tokyo (SEAMIC Publication No.62). 1991.